

WHAT IS CLAIMED:

1. A method for determining whether an analyte has a mixed androgen receptor (AR) agonist activity or an AR activity selected from the group consisting of full AR agonist activity and full AR antagonist activity, which comprises:
 - (a) providing a full-length human AR and an AR ligand binding domain polypeptide (ARLBD);
 - (b) mixing the full-length human AR with the analyte and a labeled agonist in a first binding reaction and determining a first 50% inhibitory concentration (IC₅₀) of the analyte for the full-length human AR;
 - (c) mixing the ARLBD with the analyte and the labeled agonist in a second binding reaction and determining a second IC₅₀ of the analyte for the ARLBD; and
 - (d) comparing the first IC₅₀ to the second IC₅₀ wherein a first IC₅₀ which is substantially the same as a second IC₅₀ determines that the analyte has the AR activity selected from the group consisting of the full AR agonist activity and the full AR antagonist activity and wherein a second IC₅₀ which is less than the first IC₅₀ determines that the analyte has the mixed AR agonist activity.
2. The method of Claim 1 wherein the full-length human AR and the ARLBD are each provided in a cell extract.
3. The method of Claim 1 wherein the full-length human AR is provided as an extract from human cells which endogenously express the full-length human AR.
4. The method of Claim 3 wherein the human cells are MDA-MB453 cells which have been deposited as ATCC HTB-131.
5. The method of Claim 1 wherein the ARLBD is expressed in cells selected from the group consisting of yeast cells, human cells, non-human mammalian cells, insect cells, and bacterial cells.
6. The method of Claim 1 wherein the ARLBD is a non-human ARLBD selected from the group consisting of rat ARLBD, mouse ARLBD, and rhesus monkey ARLBD.
7. The method of Claim 1 wherein the first and second binding reactions are hydroxyapatite-based displacement reactions.

8. The method of Claim 1 wherein the labeled agonist is selected from the group consisting of methyltrienolone (R1818) and 5- α -dihydrotestosterone (DHT).

5 9. The method of Claim 1 wherein the analyte is determined to have the mixed agonist activity when the second IC50 is about five-fold less than the first IC50.

10 10. The method of Claim 1 which further includes a control binding assay wherein the full-length human AR and the ARLBD are each mixed with an agonist or an antagonist and an IC50 is determined for the agonist or the antagonist.

15 11. The method of Claim 10 wherein the agonist is selected from the group consisting of methyltrienolone (R1881) and 5- α -dihydrotestosterone (DHT) and an IC50 is determined for the agonist.

12. The method of Claim 1 wherein in step (b) the analyte is labeled and the agonist is unlabeled.

20 13. A method for determining whether an analyte has an androgen receptor (AR) activity selected from the group consisting of full AR agonist activity, a full AR antagonist activity, and mixed AR agonist activity, which comprises:

(a) providing an analyte which has been determined to have the AR activity selected from the group consisting of full AR agonist activity, full AR antagonist activity, and mixed agonist activity according to the method of any one of Claims 1-12;

25 (b) providing prostate tumor cells which produce prostate-specific tumor antigen (PSA) in the presence of an AR agonist;

(c) adding the prostate tumor cells to a medium which includes an agonist to provide a first culture of the cells and adding the prostate tumor cells to a medium without the agonist to provide a second culture of the cells;

30 (d) adding the analyte of step (a) to the first culture and the second culture and incubating the cells with the analyte for a time sufficient for a control culture of the cells, which includes the agonist and not the analyte, to produce the PSA; and

35 (e) detecting the amount of PSA produced by the cells in the first culture and the second culture wherein (i) the analyte which stimulates the cells in the second culture to produce the PSA and further stimulates cells in the first culture to produce the PSA determines the analyte to have the full

AR agonist activity, (ii) the analyte which does not stimulate the cells in the second culture to produce the PSA and which reduces the amount of the PSA produced by the cells in the first culture determines the analyte to have the full AR antagonist activity, and (iii) the analyte stimulates the cell in the second culture to produce the PSA and antagonizes the ability of the agonist in the first culture to stimulate the production of the PSA determines the analyte to have the mixed agonist activity.

14. The method of Claim 13 wherein the prostate tumor cells are LnCap cells which have been deposited as ATCC CRL-1740 or CRL-1740D.

15. The method of Claim 13 wherein the amount of the PSA is detected by ELISA.

16. The method of Claim 13 wherein the agonist is selected from the group consisting of methyltrienolone (R1881) and 5- α -dihydrotestosterone (DHT).

17. A method for determining the ratio of agonist to antagonist activity of an analyte having mixed androgen receptor (AR) agonist activity, which comprises:

(a) providing an analyte which has been determined to have the mixed AR agonist activity according to the method of any one of Claims 1-12;

(b) providing prostate tumor cells which produce prostate-specific tumor antigen (PSA) in the presence of an AR agonist;

(c) adding the prostate tumor cells to a medium which includes an agonist to provide a first culture of the cells and adding the prostate tumor cells to a medium without the agonist to provide a second culture of the cells;

(d) adding the analyte of step (a) to the first culture and the second culture and incubating the cells with the analyte for a time sufficient for a control culture of the cells, which includes the agonist and not the analyte, to produce the PSA; and

(e) detecting the amount of the PSA produced by the cells in the second culture and the first culture wherein the amount of PSA produced in the second culture and the amount of PSA produced in the first culture determines the ratio of agonist to antagonist activity of the analyte.

18. The method of Claim 17 wherein the prostate tumor cells are LnCap cells which are ATCC CRL-1740 or CRL-1740D.

19. The method of Claim 17 wherein the amount of the PSA is detected by ELISA.

20. The method of Claim 16 wherein the agonist is selected from the group consisting of methyltrienolone (R1881) and 5- α -dihydrotestosterone (DHT).

21. A method for identifying an analyte which has mixed androgen receptor (AR) agonist activity, which comprises:

(a) providing a full-length human AR and a non-human AR ligand binding domain polypeptide (ARLBD);

(b) mixing the full-length human AR with the analyte and a labeled agonist in a first binding reaction and determining a first 50% inhibitory concentration (IC₅₀) of the analyte for the full-length human AR;

(c) mixing the ARLBD with the analyte and the labeled agonist in a second binding reaction and determining a second IC₅₀ of the analyte for the ARLBD; and

(d) comparing the first IC₅₀ to the second IC₅₀ wherein a wherein a second IC₅₀ which is less than the first IC₅₀ identifies the analyte as having the mixed AR agonist activity.

22. The method of Claim 21 further including determining the ratio of agonist to antagonist activity of the analyte by

(i) providing prostate tumor cells which produce prostate-specific tumor antigen (PSA) in the presence of an AR agonist;

(ii) adding the prostate tumor cells to a medium which includes an agonist to provide a first culture of the cells and adding the prostate tumor cells to a medium without the agonist to provide a second culture of the cells;

(iii) adding the analyte to the first culture and the second culture and incubating the cells with the analyte for a time sufficient for a control culture of the cells, which includes the agonist and not the analyte, to produce the PSA; and

(iv) detecting the amount of the PSA produced by the cells in the second culture and the first culture wherein the amount of PSA produced in the second culture and the amount of PSA produced in the first culture determines the ratio of agonist to antagonist activity of the analyte.